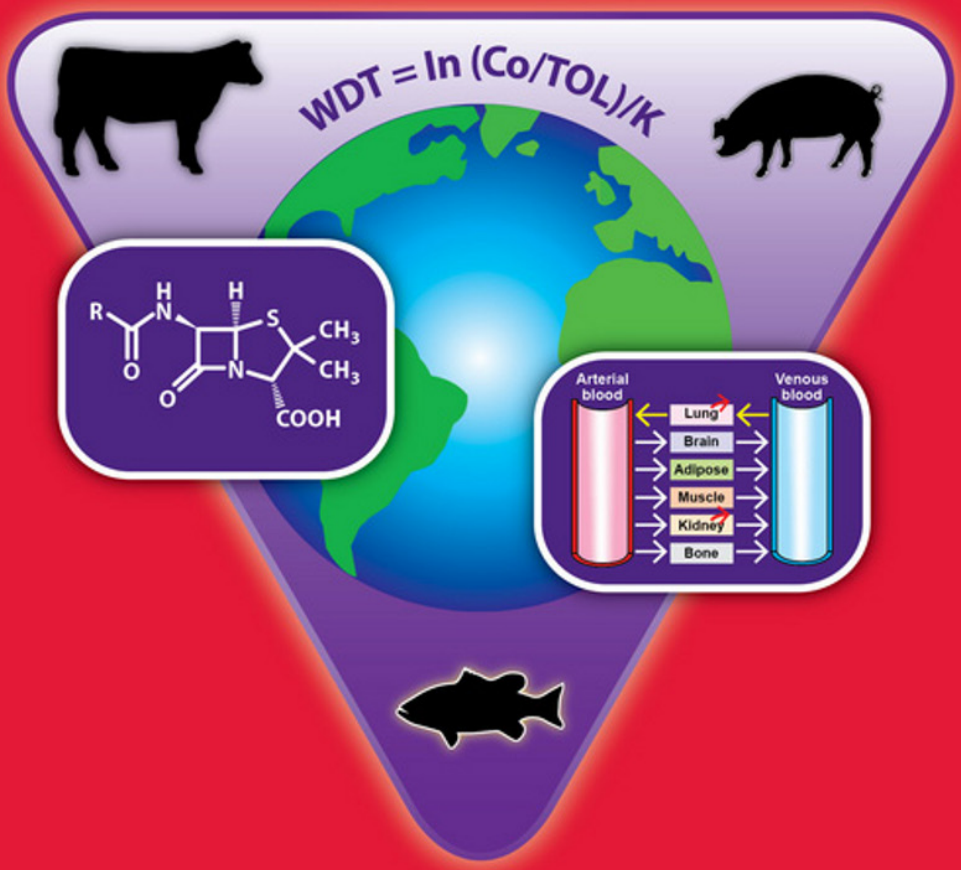
Strategies for Reducing Drug and Chemical Residues in Food Animals: International Approaches to Residue Avoidance, Management, and Testing



Edited by

Ronald E. Baynes and Jim E. Riviere

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STRATEGIES FOR REDUCING DRUG AND CHEMICAL RESIDUES IN FOOD ANIMALS

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International Approaches to Residue Avoidance, Management, and Testing

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PREFACE

The focus of this book is to present strategies that are utilized to reduce drug and chemical residues in food from livestock production, and also to present some of the newer technologies and theories that will shape how drug residues will be managed in the future. One of the novel features of this book is that it will tie in the realities of veterinary clinical practice and the use of these drugs in food animals with regulatory standards and mitigation practices.

The first half of this book focuses on strategies that are part of public policy in national and international agencies and how these agencies assess the toxicology of veterinary drugs and contaminants. This involves some discussion of how to compute safe levels (tolerances and maximum residue levels, MRLs) of these drugs and chemicals in meat and milk so that human health is not adversely affected. This section highlights the efforts at harmonization as well as differences across such jurisdictions as United States, European Union, Canada, Australia, South America, China, and Asia, where this issue has a significant impact on the trade of livestock products. This ***section also*** focuses on novel computational strategies that incorporate more statistical and mathematical approaches that are now possible with the advent of modern computers to derive safe withdrawal times. These chapters provide the reader with a general introduction to basic pharmacokinetic principles, especially those principles that are applicable in subsequent chapters in this section as it pertains to estimating a safe withdrawal time for veterinary drugs and contaminants. PK parameters and their derivation are defined in the Chapter 1. These chapters also focus on how the WDT is established in US vs. EU.

The second half of this book focuses on the use of major drug classes in livestock food animal production systems and the drugs most likely targeted for regulatory policy, pharmacokinetic modeling, and chemical residue monitoring. Each chapter in this section will be focused on subtherapeutic (feed) and therapeutic use of drugs in major livestock species such as dairy and beef cattle, swine, poultry, fish aquaculture, and small ruminant production systems. Each production system requires species-specific management practices of drug residues. Quality assurance programs are discussed for each major species with regards to species-specific management practices for controlling drug residues as well as subtherapeutic versus therapeutic drug use in livestock, and how these practices are related to the emergence of antimicrobial resistance.

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IMPORTANCE OF VETERINARY DRUG RESIDUES

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1.1 INTRODUCTION

Food animal production over the last 50–60 years has significantly increased with the implementation of modern genetics, breeding, husbandry, and nutrition. During this same time period, livestock producers have relied on the use of veterinary drugs as one of several strategies to ensure economic viability of the industry. This need for increased use of veterinary drugs, and especially antimicrobial drugs, has been linked to changes in standard livestock practices where the objective is to increase feed and space efficiency and to a need to generate greater quantities of meat, milk, and egg products in an ever increasing competitive global market. While the consumer appreciates the need to increase livestock production and generate reliable and affordable animal-derived products, this is tempered by the consumers' requirement that the food items be "free" of drugs or chemicals introduced in the production system. The wide availability of related information via the Internet has exposed the consumer to useful facts but all too often to controversial

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statements and hypotheses with very little factual support from the scientific literature regarding the prevalence of drug residues in our food, how veterinary drugs are used, and what safeguards are implemented to reduce these residues. This introductory chapter will briefly review the role of drugs in modern livestock production, quality assurance programs, adverse human health effects of drug residues, and economic impact of these residues to the livestock industry.

1.2 VETERINARY DRUG USE IN LIVESTOCK

Modern livestock production can be described as involving intensive animal production practices that often use veterinary drugs at subtherapeutic level in feed and water in order to improve feed efficiency for growth and production and maintain animal health. In such close animal–animal contact practices, prevention of disease is more important than treating for disease that would require therapeutic levels (higher doses) of the drug. The United States defines subtherapeutic use of an antimicrobial as a feed additive less than 200 g of drug per ton of feed.

Subtherapeutic drug use may take the form of (i) *antimicrobials* delivered to the animal as a feed or water additive and (ii) *hormones* delivered via ear implants or feed additives.

The antimicrobials approved in the United States and EU to be used in this legal manner often belong to the tetracycline, sulfonamide, or macrolide class of antimicrobials. Several EU countries and others banned or limited the use of these drugs as growth promoters as there are concerns that their use promotes the emergence of antimicrobial resistance. This cause-and-effect relationship is continually being debated across various jurisdictions; although epidemiological evidence continues to accumulate, definitive conclusions from rigorous research in livestock production systems has not been forthcoming. This issue will be further explored in this and other chapters of this book.

The use of hormone growth promoters in livestock has also been a controversial debate as various regulatory authorities in different jurisdictions regulate these drugs in a different manner. The U.S. FDA has approved the legal use of 17β-estradiol, testosterone, progesterone, trenbolone, and zeranol as solid ear implants and melengestrol acetate (feedlot heifers) and ractopamine (swine) as feed additives. Compared to the United States, the EU in 1988 issued a total ban of all hormonal active growth promoters in livestock production. Prior to 1988, in the Netherlands (1961) and Belgium (1962–1969), there was a total ban on anabolic agents for growth promotion purposes in slaughter animals in order to protect consumers and for the benefit of international trade (Stephany, 2010). It should be noted that the

United States challenged the EU's ban, and in 1998, the WTO found that the EU's ban was not supported by science and inconsistent with WTO obligations (USTR, 2009).

Therapeutic drug use in veterinary livestock involves administration of veterinary drugs according to label to treat an individual animal or herd or flock of animals by various approved routes of administration. The use of water additives is recognized in all countries as a form of therapeutic drug use and not subtherapeutic drug use or for growth promotion purposes. It has however been our experience (Mason et al., 2012) that treatment of large herds via water medication does not always result in each animal in the herd receiving therapeutic drug levels. This has often been associated with competition between animals in the herd and/or malfunctioning medicators. The approved use of the many therapeutic drugs will be outlined in the species-specific chapters of this book.

The passage of the Animal Medicinal Drug Use Clarification Act (AMDUCA) 1994 in the United States allows food animal veterinarians to administer drugs in an extralabel manner within certain guidelines as outlined in the following text. Veterinarians often have to resort to using these drugs in an extralabel manner for a number of reasons. *New generics of old drugs are approved based on bioequivalence to pioneer formulation*, which allows the same dosage and milk discard/meat withdrawal times. The problem with this approach is that new bacteria being treated have much higher MICs than bacteria and microorganism many years ago, and thus, higher dose must now be used. Veterinarians often consult with FARAD to find out new withdrawal times, and this is described in more detail in Chapter 14. The scientific issue is that most antimicrobials used in dairy practice today are old drugs (or generic copies of old drugs) that are now not effective unless given at higher doses, necessitating extended milk discard times. Risk of exposure to low-level residues of most other drugs out there is "theoretical," but low label dosages of antimicrobials, used to insure adequate withdrawal times, will promote resistance, which is the major public health issue. There are more modern approaches that would allow dosage adjustments with new withdrawal times, but we are stuck in the science of the 1970s. *Legal precedence and business issues tend to hand tie the FDA* (in approving all generics just like the first one that was approved even if science has advanced in 30 years). Production use of antibiotics as growth promoters may very well be banned, and therapeutic use at higher doses by licensed vets maintained.

Phytoceuticals are increasingly being used on organic farms with varying degrees of success. These drugs are not regulated by the FDA-CVM as they are often described as "generally recognized as safe" (GRAS). There are however several guidance documents and requirements that organic livestock farms are required to follow and are discussed elsewhere in this book.

1.3 QUALITY ASSURANCE PROGRAMS

Consumers are very aware of drug and chemical use in the livestock industry, and oftentimes, there is general misinformation about how these drugs are used in the industry. The infrequent catastrophic drug residue violations are often a direct result of careless farm management. *The subsequent economic cost to the livestock industry is not ignored by the many stakeholders involved in livestock production and distribution and sales of meat, milk, fish, and egg products*. This will be discussed in more detail in a later section of this chapter.

In lieu of these scenarios, the livestock industry has been aggressively policing itself to make sure that producers are educated and trained to prevent drug residue violations on their farms. Many if not most livestock producers follow and adhere to their respective quality assurance programs for their commodity group that attempt to minimize drug residue violations and promote judicious use of veterinary drugs. A summary of the steps producers are encouraged to follow whether it is the beef, dairy, pig, goat, or poultry industry is as follows:

1. Improve husbandry practices by maintaining appropriate husbandry, hygiene, examinations, and vaccinations.
2. Consult with a veterinarian prior to use of drugs or medicated feed or water as therapeutic alternatives may be more appropriate.
3. Use drug according to veterinary label and only resort to using veterinary drugs as a last resort. This is especially important for antimicrobial drug use.
4. Antimicrobial drug use is inappropriate for viral infections without bacterial complication.
5. Optimize antimicrobial drug regimen using current pharmacological information and principles.
6. Mitigate veterinary drug spillage into the environment.
7. Keep good records of drug use on each farm.
8. Extralabel drug use in the United States must follow the FDA regulations: prescriptions, including extralabel use of medications must meet the AMDUCA amendments to the Food, Drug, and Cosmetic Act and its regulations. This includes having a valid veterinary–client relationship.

The passage of the AMDUCA in the United States in 1994 allows food animal veterinarians in the United States to administer drugs in an extralabel manner within certain guidelines. Several chapters in this book will focus on

PK principles that can be used to extrapolate across and within species, across routes of administration, and across doses. To date, legislation similar to AMDUCA does not exist in other major livestock-producing countries.

1.4 ADVERSE HUMAN HEALTH EFFECTS OF DRUG RESIDUES

Inappropriate use of several of veterinary and human drugs in livestock production can result in significant residue levels in meat, dairy, and poultry products that can cause adverse health effects in consumers. Although approximately 80% of all food animals are given drugs during their lifetime, residue violations are often less than 1% thanks to rigorous surveillance and testing in major livestock-producing countries and increasing so in smaller developing states. However, many consumers in developed and developing states rely on livestock products as their major source of protein. The average American consumes 200 pounds of meat and fish, 67 pounds of poultry, 30 pounds of eggs, and 600 pounds of dairy products annually. In spite of the low level of drug residue contamination, this high level of consumption of livestock products increases the possibility that any one violative incident can result in adverse health effects affecting more than one individual or community following acute or chronic exposure.

A very good example of the aforementioned case was associated with clenbuterol residues. In one 6-month period in 1993, more than 1200 hospitalizations and 3 deaths in France and Spain were reported to have resulted from eating beef livers contaminated with clenbuterol. One study documented in Portugal four cases of acute food poisoning, involving a total of 50 people, due to the ingestion of lamb and bovine meat containing residues of clenbuterol (Barbosa et al., 2005). An outbreak with hospitalization was described in Italy in 1997 involving 15 people within 0.5–3.0 h after the consumption of veal and not livers (Brambilla et al., 2000). No deaths were reported but clinical signs and symptoms disappeared within 3–5 days. More recently, 286 villagers in Changsha, capital of Hunan province in China, were hospitalized and suspected to have been made sick from consuming clenbuterol-tainted pork (UPI, 2011). Symptoms of clenbuterol intoxication can be described as predominantly gross tremors of the extremities, tachycardia, nausea, headaches, and dizziness. This drug is a beta-agonist, acts as a bronchodilator, and can have anabolic effects such as increase lean body mass and weight gain. It is not approved for use in humans or in food animals by the U.S. FDA, and extralabel use in food animals is strongly prohibited. However, there is approval for use in horses with recurrent airway obstruction (heaves), and there are no studies to support meat withdrawal times for this drug given to horses intended for food.

1.5 WITHDRAWAL TIME DETERMINATIONS

Several chapters in this book will describe in brief several of the methods used by the U.S. FDA (2006) and the European Medicinal Agency (EMA, 1996, 2000) to derive regulatory withdrawal times that ensures the consumer is protected from exposure to drug concentration that will cause adverse health effects. The guidance documents for these calculations from each of these regulatory authorities are always changing with new revisions, and they may vary slightly, but there are some common features that the reader should appreciate.

For example, in assigning a milk withdrawal time, the U.S. FDA uses an algorithm that calculates the upper 99th percentile of the population and 95th percent confidence limit. As with the tissue withdrawal period, this assures that when the drug product is used according to its approved label, there is only a 5% chance that one animal in 100 will have milk residues above the milk tolerance concentration. In the EU, the recommended method is also a statistical method based on a linear regression model in which the upper 95% tolerance limit of the 95% percentile of the residue depletion curve is used to determine the withdrawal period. As per the U.S. FDA, the minimum number of animals in a milk residue study is 20, based on the statistical requirements for the calculation of the withdrawal time. In the EU, milk withdrawal periods are established for individual animals and not for tank milk as per the U.S. FDA-CVM. The reader is encouraged to consult with updated guidance documents in the respective jurisdictions with regard to recommended regulatory methods to calculate the meat and milk withdrawal times. There are several chapters in this book that describe alternative and more flexible pharmacometric methods that utilizes the current advances in mathematical modeling and well-accepted software that considers a larger population of animals and other variables such as production and disease status that are often overlooked in the current regulatory methods in many jurisdictions. These novel methods are not currently accepted by regulatory agencies in the establishment of meat and milk withdrawal periods for veterinary drugs. However, several of them such as physiologically based pharmacokinetic (PBPK) modeling have been adapted with success by the U.S. EPA in their guidance for conducting a human health risk assessment of environmental contaminants.

1.6 ANTIMICROBIAL RESISTANCE

The U.S. FDA in 2010 provided guidance on the judicious use of antimicrobial drugs in livestock and recognized that failure of antimicrobial therapies in humans can be related to human and animal use of antimicrobials among

other factors. The FDA believes that "the use of medically important antimicrobial drugs in food-producing animals for production purposes (e.g., to promote growth or improve feed efficiency) represents an injudicious use of these important drugs. **Production uses are not directed at any specifically identified disease, but rather are expressly indicated and used for the purpose of enhancing the production of animal-derived products**. In contrast, FDA considers uses that are associated with the treatment, control, or prevention of specific diseases, including administration through feed and water, to be uses that are necessary for assuring the health of food-producing animals." This topic is discussed in more detail in subsequent chapters of this book that describe the prudent drug use of antimicrobials in ruminant and pig production systems.

1.7 ECONOMIC IMPACT OF DRUG RESIDUES

There is a significant economic impact associated with drug residues in meat, milk, or egg products. Besides loss in sales of product, public perception can have the greatest impact on consumers already weary about drug and chemical use in food production systems in developing and developing countries. Oftentimes, the consumer is exposed to misinformation from media sources whose understanding are limited with regard to how these drugs are used on livestock farms and the many stages between the farm and table where residue violations are prevented. The remainder of this book will highlight many of the established practices that are effective in the mitigation of drug residues and scenarios where residue violations are likely to occur and warrant future research and attention by regulatory authorities.

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2

PHARMACOKINETIC PRINCIPLES FOR UNDERSTANDING DRUG DEPLETION AS A BASIS FOR DETERMINATION OF WITHDRAWAL PERIODS FOR ANIMAL DRUGS

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2.1 INTRODUCTION

According to the U.S. Food and Drug Administration's (FDA) Center for Veterinary Medicine (CVM), an approved animal drug is considered to be safe and effective, if it is used according to its label instruction—safe for use in the intended species as well as for human consumption of the edible products derived from animals treated with the drug. An evaluation of drug safety for human consumption includes an assessment of toxicology and residue chemistry—as described in "FDA CVM's Guidance for Industry (GFI) #3: General Principles for Evaluating the Safety of Compounds Used in Food Producing Animals (FDA GFI #3, 2006)," and all the toxicology-related GFIs. In addition, the human food safety evaluation for active pharmaceutical ingredients (API) possessing antimicrobial activity also includes an assessment of the effect of the transmission of food-borne bacteria of human health

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concern through the consumption of animal-derived food products (FDA GFI #152, 2003) and an evaluation of the safety of drug residues with respect to the human intestinal flora for establishing a microbiological acceptable daily intake (FDA GFI #159, 2011).

The use of approved drugs in food-producing animals can lead to the presence of unsafe drug residues in the edible products above the established tolerances (21 CFR 556) if drugs are not used according to their label directions (i.e., if animals are sent to slaughter before the established withdrawal period has been observed). Brynes (2005) provided a more recent review on the history of tolerances for residues of new animal drugs in food. In addition to providing updated definitions and concepts of establishing and promulgating regulations on tolerances, Brynes provides a historical perspective on how the establishment of residues changed over time (e.g., the earliest tolerances generally referred to the parent drug, which was later changed to establish tolerances that would reflect the total residue). The presence of violative drug residues in food may result in potential risks to humans consuming residues, including acute and chronic toxicities, such as allergic reactions, various adverse reproductive and developmental effects, carcinogenicity, as well as a risk of the development of antimicrobial resistance (Horrigan et al., 2002). The edible products considered in the human food safety evaluation include muscle, liver, kidney, skin with or without fat, and milk and eggs (when appropriate). Residues of human food safety concern may include the APIs and excipient(s) of a drug product, drug metabolites, and any substance formed in or on the edible food products as a result of drug treatment.

Risk assessment principles based on the standard of reasonable certainty of no harm for human consumption are applied in the human food safety evaluation of animal drug residues in food animals. The assessment comprises an evaluation of the traditional toxicological effects of drug residues on human health, the amount of residues human consumers are exposed to, the risk of developing antimicrobial drug–resistant bacteria due to the use of antimicrobial drugs in animals, and the effects of drug residues on the human intestinal flora (Friedlander et al., 1999). It should be emphasized that multiple and robust layers of safety are factored in during the risk assessment process for the evaluation of human food safety of new animal drugs to accommodate various intrinsic (i.e., related to the animal physiology) and extrinsic factors (i.e., factors influencing the drug's characteristics, food, environment, concomitant medications, etc.) that can influence a drug's behavior in the body. In this chapter, a brief summary of basic pharmacokinetic (PK) principles is provided to help readers understand the pharmacologic principles underlying the human food safety evaluation of new animal drugs.

The depletion of residues of a compound may vary considerably due to the impact of various intrinsic and extrinsic factors, as described below.

Total residue evaluation consists of evaluation of the parent compound, free metabolites, and metabolites that are covalently bound to endogenous molecules. The levels of residues depend on the types of tissues, the amount of the drug administered, and the time following the last drug administration to the animal (FDA GFI #3, 2006). Therefore, FDA typically requires that residue chemistry studies be conducted in each species/class of animal for which the sponsor is seeking approval. Studies also may be needed in specific classes of animals (e.g., veal calves, lactating dairy cattle for milk, laying hens for eggs). The studies are typically conducted in a limited number of animals in order to minimize the economic and ethical impacts on new animal drug sponsors (Martinez et al., 2000). However, it is critical that the human food safety evaluation is conducted in the appropriate population of animals for which the drug is intended. The human food safety studies use the highest intended treatment dose, the longest intended treatment duration (or a duration that ensures that the drug concentrations have reached steady state), and the intended administration route (and therefore represent the worst-case scenario in terms of drug residue exposure to humans). Because of these conditions, the human food safety studies for new animal drug approvals are typically conducted once the dose and dosing regimen of the drug have been firmly established and once when the sponsor has identified the final formulation for their new animal drug product. Depending on the dosing regimen, the design of required studies may differ considerably. For example, if a drug is intended to be administered once for a specific therapeutic effect, then a single dose of drug in the target animals will capture appropriate exposure; on the other hand, if a drug is intended for prolonged treatment, it is critical to evaluate residue depletion after the drug concentrations have reached the steady state.

Before discussing specific study designs for various kinds of residue depletion studies (which will be covered in Chapter 3), it is important to understand the pharmacologic basis for recommending those study designs. This chapter reviews the impact of various internal (endogenous) factors on *in vivo* drug behavior, which includes both blood and tissue levels. The withdrawal time, a critical factor for ensuring human food safety, is in essence a PK parameter based on the legal target tissue tolerance and reflecting the drug's rate of depletion from that target tissue (Riviere, 1999).

The amount of drug substances in edible animal products is a complex function of the rate and extent of absorption of the parent compound, the formation of metabolites (free and covalently bound to endogenous molecules), and the distribution and clearance of the parent compound and its metabolites. Drug distribution depends on the physicochemical properties of the drug, the concentration gradient between the blood and tissue, the ratio of the blood flow to tissue mass, and the affinity of the drug for tissue (Riviere, 1999).

Tissue depletion reflects the drug's partitioning characteristics between blood and tissue, the blood flow to that tissue, and the rate at which the drug is depleted from the systemic circulation. In some instances, the tissue itself (especially the liver and kidney) may also be involved in drug metabolism, which then further contributes to the overall tissue rate of depletion. Therefore, drug residues in the various edible products will deplete at different rates, and their respective tissue elimination half-lives have to be determined for the establishment of the withdrawal time. The final withdrawal period assignment is based upon the time it takes for the marker residue to deplete from the slowest depleting tissue (the target tissue).

2.2 BASIC PHARMACOKINETIC PRINCIPLES UNDERLYING DRUG DEPLETION

For any desired level of drug exposure (typically expressed in terms of an area under the concentration versus time curve, AUC), critical points to consider are the dose administered (D), total body clearance (Cl), and bioavailability (F). Drug exposure is determined by the following equation:

$$\mathrm{AUC}_{0-\mathrm{inf}} = \frac{\mathrm{D} \times \mathrm{F}}{\mathrm{Cl}} \tag{2.1}$$

It is important to note that the targeted dose is equally influenced by the Cl and F. Bioavailability (or fraction of administered dose that is absorbed) is the proportion of the administered dose that reaches the systemic circulation. It is a function of animal physiology, route of administration, and the physicochemical characteristics of the API and the formulation (Martinez and Amidon, 2002). Clearance represents the volume of whole blood, serum, or plasma completely cleared of drug per unit of time. Unlike the F, Cl is solely a function of the physicochemical properties of the API and the host physiology (unless a specific ingredient interacts with the elimination process).

All aspects of the PK response (absorption, distribution, metabolism, and elimination (ADME)) are important in understanding the human food safety effects, as they can ultimately influence drug depletion profiles. Although not a critical factor for immediate-release drugs, absorption can significantly affect depletion times of modified release dosage forms, due to the presence of flip-flop kinetics (where the rate of drug absorption rather than elimination is the rate-limiting factor determining the slope of the terminal phase of the concentration versus time profile).

Distribution of a drug to peripheral tissues is affected by the binding of the drug to blood and tissue macromolecules, blood flow, partition coefficient of the drug between the blood and the organs into which it distributes, and the physicochemical properties of the drug. Tissue binding, which tends to increase drug distribution, is an important underlying consideration in the evaluation of the human food safety of edible products derived from animals treated with a new animal drug. The tissue drug concentrations determine the time needed for drug-related residues to deplete to legally established tissue tolerances, which are, in turn, based upon the safety of the residues to humans consuming edible products of animals treated with a new animal drug, extrapolated from studies in toxicological model species.

The drug distribution between plasma and tissues is described by the PK parameter, the volume of distribution (Vd). Vd is not a physiologic value, but rather a reflection of how a drug gets distributed throughout the body, the latter depending on its physicochemical properties, such as solubility, charge, and size. Drugs that remain in the circulation tend to have a low Vd, whereas drugs that are highly bound to tissue tend to have a very high Vd. Vd relates the mass of drug in a compartment to the volume into which it is diluted and is described by the following equation:

$$\text{Vd} = \frac{\text{Dose}}{\text{Drug plasma concentration}} \tag{2.2}$$

The term Vd may be expressed as either Vd_c, Vd_{area}, Vd_{ss}, or Vd_{β}. Volume of distribution of the central compartment, Vd_c, reflects the volume of the central compartment, before any distribution has taken place, and relates the dose to the drug concentration at time 0.

The apparent volume of distribution, Vd_{area}, is based on the total AUC. It relates plasma concentration to the amount of drug in the body at all times after distribution equilibrium is reached after a single dose or multiple discrete doses. It is calculated as follows:

$$\text{Vd}_{area} = \frac{\text{Dose} \times \text{F}}{\text{AUC} \times \beta}, \tag{2.3}$$

where β is the slope of the terminal portion of the plasma concentration–time curve (plotted as a natural logarithm of concentration versus time).

The volume of distribution at steady-state, Vd_{ss}, provides an estimate of drug distribution independent of elimination processes, which is most useful for predicting the plasma concentrations at steady state. Steady state is reached when the free concentration of drug in the plasma equals the free

concentration in the tissue. It is a correct measure for continuous intravenous infusion or at a single instant in time (when the rate of elimination equals that of distribution) and is calculated as:

$$\mathrm{Vd}_{ss} = \frac{\mathrm{Dose} \times \mathrm{F} \times \mathrm{AUMC}}{\mathrm{AUC}^2}, \tag{2.4}$$

where AUMC is the area under the moment curve, which is the integral of the curve plotting the product of concentration and time by the time the concentration was observed.

The apparent volume of distribution in the postdistribution (or terminal) phase, Vd_{β}, neglects the distribution phase of drug disposition and is calculated as follows:

$$\mathrm{Vd}_{\beta} = \frac{\mathrm{Dose} \times \mathrm{F}}{B}, \tag{2.5}$$

where B is a value obtained from extrapolating the linear terminal portion of the plasma concentration–time curve to its intercept on the y axis (plasma drug concentration).

Because the Vd_{β} ignores the distribution phase, it is valid only for drugs that fit a one-compartment model (it generally overestimates the true volume of distribution of multi-compartmental drugs). The only measure of volume that is independent of the rate of chemical elimination is the Vd_{ss}.

V_d may also be used to determine how readily a drug will displace into the body tissue compartments relative to the blood using the following equation:

$$V_d = V_p + V_t\left(\frac{f_u}{f_{ut}}\right), \tag{2.6}$$

where V_p is the plasma volume, V_t is the apparent tissue volume, f_u is the fraction unbound (free) in plasma, and f_{ut} is the fraction unbound (free) in tissue.

Understanding the drug distribution and the presence of peripheral compartments is important when evaluating drug depletion from the body. A drug that selectively binds to tissues or sequestered into a deep compartment may have several different half-lives, and it is critically important to understand drug depletion for the determination of the withdrawal time. In addition, the analytical method has to be sufficiently robust and sensitive to address the tissue distribution of a drug and potential presence of deep peripheral compartments, which could result in "spikes" in residue concentrations above the tolerance.

As the drug is absorbed and distributed throughout the body, drug elimination becomes the most predominant process. Mechanisms of drug elimination include biotransformation (metabolism) and excretion. In general, both mechanisms are involved in drug elimination, although one mechanism is usually dominant over the other. Of the physicochemical properties that determine the mechanism of elimination, lipid solubility and degree of ionization seem to play the most critical role. For example, lipid-soluble drugs undergo biotransformation by hepatic microsomal enzymes, while many polar drugs and metabolites are excreted by the kidney (Brown, 2001).

Most commonly, a constant proportion of the dose is cleared over time, which is termed first-order elimination. By definition, in first-order or linear processes, the elimination rate (ke) is constant, while the actual rate of the process varies in direct proportion to the dose. The concentration (C) at any time (t) after a single intravenous dose administered can be calculated as:

$$Ct = \left[\frac{\text{Dose}}{V_{\text{d}}}\right] \times e^{-\text{ke}\times t}, \tag{2.7}$$

where e is the base of the natural logarithm ($e = 2.713$), and the elimination constant (ke) is represented by the ratio of clearance to volume of distribution and is usually expressed in units of 1/h.

$$\text{ke} = \frac{\text{Cl}}{V_{\text{d}}} \tag{2.8}$$

Most equations in this chapter describe a one-compartment body model with no absorption. This is an oversimplified description of the PK processes, because most drugs are not adequately described by a one-compartment body model: the body does not behave as a single homogeneous compartment and there is usually no instantaneous distribution through this one compartment. These one-compartment equations are included as illustrations of the principles used in PK, but the reader should keep in mind that for most drugs the body does not behave as a single compartment and that the understanding of multiple compartments is critical for understanding drug depletion and the risks for violative residues in tissues.

In a multi-compartmental model, different body compartments are characterized by different rates of drug distribution. Most typically, there are two major body compartments, although there can be more than two, depending on the rates of drug distribution among the compartments. A two-compartment body model is schematically represented in Figure 2.1. It consists of a central compartment, which comprises blood plasma and the extracellular fluid of

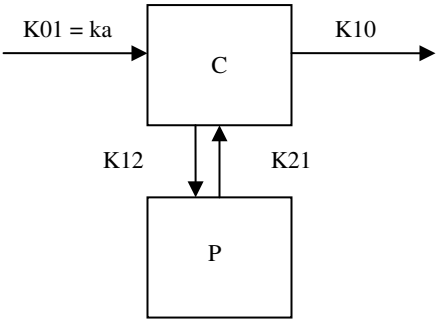

FIGURE 2.1 A two-compartment body model with first-order absorption.

highly perfused organs (such as the heart, lungs, kidney, and liver), and a peripheral compartment, in which the distribution occurs more slowly (such as in muscle and fat). In addition to the presence of multiple compartments, many drugs are administered extravascularly, so there is an absorption phase that needs to be taken into consideration when modeling the drug PK response (illustrated by the $k01 = \text{ka}$ arrow).

Figure 2.1 shows a two-compartment model with a first-order absorption, where C is the central compartment (1), P is a peripheral compartment (2), $k01$ is the absorption rate constant, $k10$ is the elimination rate constant, and $k12$ and $k21$ are the inter-compartmental constants reflecting distribution.

For a drug administered extravascularly and assuming a non-instantaneous distribution (e.g., a two-compartment model), the plasma concentration at any time can be calculated as:

$$Ct = A \times e^{-\alpha \times t} + B \times e^{-\lambda z \times t} - C \times e^{-\text{ka} \times t}, \tag{2.9}$$

where A, B, and C are the y-axis intercepts for the slopes α (rapid redistribution phase), λz (elimination phase), and ka (absorption phase), respectively.

Elimination rate for a two-compartment model is calculated as follows:

$$\text{ke} = \frac{k10 \times k21}{\left(k21 + k12\right)} \tag{2.10}$$

Absorption rate of a drug is determined by the slope of the relationship between the logarithm of the amount of the drug absorbed and time. In first-order absorption, a constant fraction of the drug is absorbed per unit of time and the absorption process is thus linear. In contrast, if the saturation of the absorption mechanism occurs, the process may become nonlinear due to capacity limitations (with a lower percent of the dose absorbed at higher doses).